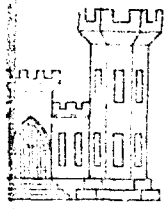

DREDGED MATERIAL RESEARCH PROGRAM

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BIOASSESSMENT OF THE STANDARD ELUTRIATE TEST

by

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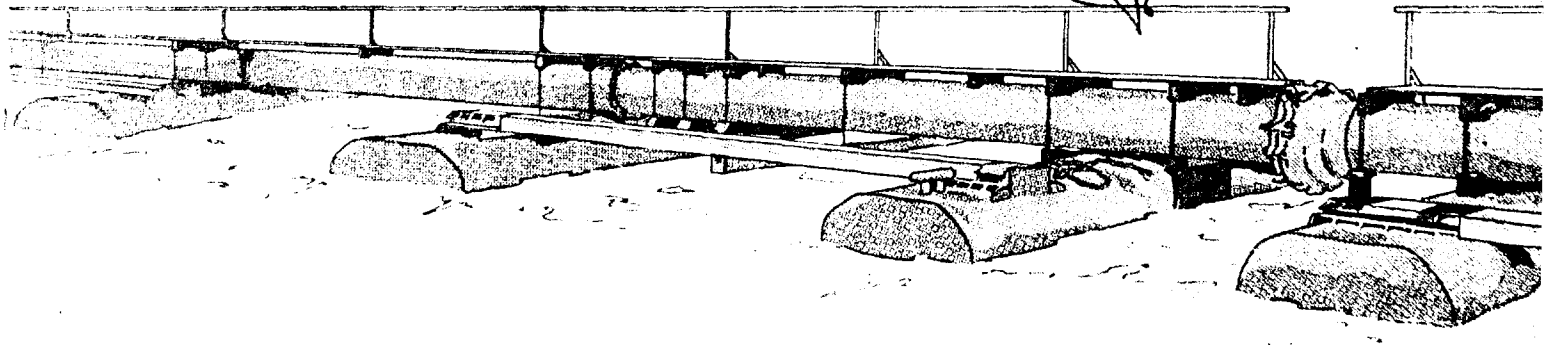

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Biological assessment studies of the standard elutriate have been conducted using selected species of algae, bacteria, and protozoans as representative test organisms. Algal bioassays have proven useful in providing information about the biologically available nutrients released from the sediments. Growth was determined by measuring the maximum number of cells per milliliter of treatment. The treatments included 100-percent disposal site water, 100-percent elutriate, and various combinations of elutriate and disposal site water. Elutriates prepared from some locations stimulated algal growth when (Continued)

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compared with the growth obtained in the disposal site water. Other elutriate preparations demonstrated an inhibitory effect toward growth of the test algae. The algal assay procedure is a useful method for evaluating potential effects of dredging and dredged material disposal on phytoplankton at the proposed discharge site.

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PREFACE

This paper was prepared for the American Society of Civil Engineers Specialty Conference on Dredging and Its Environmental Effects and was presented in Mobile, Alabama, on 27 January 1976.

The work described herein was conducted under Task Area 1E, Pollution Status of Dredged Material, of the Dredged Material Research Program (DMRP), at the U. S. Army Engineer Waterways Experiment Station (WES), Vicksburg, Mississippi. The task area is part of the Environmental Impacts and Criteria Development Project, Dr. Robert M. Engler, Manager.

The paper was prepared by Dr. Peter J. Shuba, Dr. Henry E. Tatem, and Mr. Joe H. Carroll. The paper was presented by Dr. Shuba. The report was prepared under the general supervision of Dr. John Harrison, Chief, Environmental Effects Laboratory (EEL), and Dr. Rex L. Eley, Chief, Ecosystem Research and Simulation Division, EEL.

Directors of WES during preparation and publication of this report were COL G. H. Hilt, CE, and COL J. L. Cannon, CE. Technical Director was Mr. F. R. Brown.

CONTENTS

BIOASSESSMENT OF THE STANDARD ELUTRIATE TEST

By Peter J. Shuba,[1] Joe H. Carroll,[2] and Henry E. Tatem[3]

INTRODUCTION

An area of major concern to the Dredged Material Research Program (DMRP) is the immediate effect of chemicals released from the suspended dredged sediments on water quality and aquatic ecology during dredging and disposal operations. To address this concern, the biological assessment work unit was established to develop techniques useful in interpreting the standard elutriate test. The elutriate test provides a measure of the change in concentration of certain contaminants at the disposal site. Information that relates the release of these chemicals to their effect on biota is lacking. Specific objectives of the research were to determine the biological effects of the soluble chemicals released from sediments during dredging and disposal operations. Chemical analyses of the elutriate defined the concentration of selected nutrients and heavy metals. Biological assessment documented the response of selected test organisms to the elutriate. Correlations between chemical composition and biological response could be of value in establishing disposal criteria for dredged material.

Bioassay has been defined as "any test in which organisms are used

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to detect or measure the presence or effect of one or more substances or conditions" (4). Alderdice (3) stated there are three parts to a bioassay: (a) a stimulus, such as a drug, insecticide, or industrial waste; (b) a subject which may be a cell, a tissue, or a total organism; and (c) the subject's response.

Since microbes are abundant and ubiquitous in aquatic environments (6), it is possible that an inhibitory or stimulatory effect on one or more of their biological functions would provide information useful in predicting an effect on other aspects of the ecosystem. Therefore, it was of interest to predict the effect of soluble chemicals released from the sediment on microbial communities at the disposal site. Representative species of microorganisms were selected as test organisms to serve as biological indicators in the development of analytical techniques.

Microorganisms are important members of aquatic ecosystems. Algae are primary producers converting carbon dioxide to organic cell material which is introduced into the food chain when algae are used as food by higher trophic levels, or upon their death and decomposition. They produce large quantities of oxygen for use in respiration by members of the ecosystem, and algal blooms are problems in many water supplies because of smell or taste which they impart to the water (2). Procedures employing algae to assess the nutrient status of fresh and salt water are established and generally accepted (1,9).

The bioassay work using sediments and water samples from Ashtabula Harbor was started during the last week in July 1975 to coincide with the beginning of long-term field studies in the area including the investigation of planktonic communities, benthic assemblages, fish populations, water quality parameters, physico-chemical sediment parameters, hydraulic regime and physical nature of the bottom sediments associated with the Ashtabula Harbor disposal site. A sediment survey of Ashtabula Harbor was conducted during February 1975 by the U. S. Environmental Protection Agency (EPA). Based on the results of bulk sediment analysis and the standard elutriate test, a portior of the harbor was designated polluted while another portion was assigned a nonpolluted status. One objective of the bioassessment work on Ashtabula samples was to

determine if the polluted sediments would produce significantly different effects on algal growth from those of nonpolluted sediments taken from the same harbor. A second objective was to compare the growth of the test species in sediment elutriates, slurries taken from the hopper during the dredging operation, and water collected from the disposal plume during the disposal operation.

Houston Ship Channel sediments were selected because the work unit team was searching for some heavily polluted sediments. Houston sediments have been reported to contain high concentrations of heavy metals and petroleum hydrocarbons. It was anticipated that some of these contaminants would be released from the sediment during elutriate preparation and would result in a dramatic effect upon algal growth.

METHODS

Sampling Procedures

Sediment samples were obtained using an Ekman dredge. Water samples were obtained using a Van Dorn water sampler. Sediment and water samples were immediately placed into 1-gal (3.785-ℓ) polyethylene jugs and placed in a cooler of ice. Hopper samples were collected in plastic jugs. The samples were kept on ice until they were returned to the laboratory and prepared for bioassay studies.

Preparation of Elutriate

Elutriates were prepared using a modification of the technique described by Keeley and Engler (7). Three hundred ml of unfiltered dredge site water was placed in a 1-ℓ flask, and 100 ml of sediment was added by displacement of the liquid volume. Final volume was brought to 500 ml with dredge site water. The flasks were placed on a wrist-action shaker for 30 min of vigorous shaking. After a 1-hr settling period, the contents of the flasks was poured into 1-ℓ plastic centrifuge bottles and centrifuged at 6000 rpm for 10 min. The supernatant was then filtered through 0.45-μm pore size millipore filters. The disposal site water used to dilute the elutriates was filtered in the same manner as were the hopper slurries and plume samples.

Chemical Analyses

Disposal site water, dredge site water, and elutriates were analyzed by the Analytical Laboratory Group (ALG) of the Environmental Effects Laboratory (EEL) at the Waterways Experiment Station (WES). Procedures and methods for chemical analyses were those described in "Methods for Chemical Analysis of Water and Wastes" (10) and "Standard Methods for the Examination of Water and Wastewater" (15). Nutrient analyses included ammonia plus ammonium (NH_3+NH_4^+), nitrate- (NO_3) and nitrite-nitrogen (NO_2), orthophosphate (OPO_4), acid-hydrolyzable phosphate ($AHPO_4$), total organic carbon (TOC), total inorganic carbon (TIC), and total Kjeldahl nitrogen (TKN). Heavy metal concentrations were determined for cadmium (Cd), nickel (NI), zinc (Zn), manganese (Mn), lead (Pb), coppper (Cu), iron (Fe), and arsenic (As).

Algal Assay Procedures

The algal assays consisted of establishing a series of treatments and controls using elutriate and filtered disposal site water. These experimental units were inoculated with a test organism taken from a stock culture and held under a specified set of test conditions while a sampling program was conducted to evaluate potential effects. The algal assays for freshwater dredging and disposal sites were based on the procedures described in "Algal Assay Procedure: Bottle Test" (1). The assays for marine and estuarine dredging and disposal sites followed the procedures described in "Marine Algal Assay Procedure: Bottle Test" (9).

Selenastrum capricornutum was selected as the test alga for freshwater biological assessment studies, and Dunaliella tertiolecta was used as a representative marine alga. Stock cultures of both organisms were obtained from the EPA's National Environmental Research Center, Corvallis, Oregon. Selenastrum capricornutum is a unicellular or loosely aggregated colonial green algae, Class Chlorophyceae, Order Chloroccocales. Individual Selenastrum cells are curved and range in size from 20 to 48 µm in length and from 3 to 9 µm in width. Dunaliella tertiolecta is a green unicellular flagellate, Class Chlorophyceae, Order Volvocales. Cells are ovoid and attain a size of

5 to 8 by 10 to 12 µm with two long flagella at the anterior end.

Stock algal cultures were grown in synthetic nutrient medium (1,9). Fresh cultures were started once a week by transferring 0.1 ml of a 1-week-old culture to 100 ml of fresh medium using aseptic techniques. Stock cultures were grown at laboratory temperature (approximately 23°C) under continuous cool-white fluorescent lighting at an intensity of approximately 1500 µw/cm^2 while being shaken continuously at 110 rpm.

Culture vessels were 500-ml Pyrex Erlenmeyer flasks stoppered with polyurethane foam plugs. All glassware was washed with detergent, rinsed with tap water, placed in a 10-percent hydrochloric acid bath for a few hours, and rinsed five times with tap water and five times with distilled water.

Treatment levels were established using dredged material elutriate, disposal site water, and an inoculum of the test organisms in 500-ml Erlenmeyer flasks with a total liquid volume of 100 ml. The following treatment levels were used:

Percent Elutriate	Percent Disposal Site Water
0	100
25	75
50	50
75	25
100	0

Controls included viability checks using 10- and 100-percent synthetic algal nutrient media. Also, 100-percent disposal site water, 100-percent elutriate, and a 50-percent disposal site water to 50-percent elutriate mixture received an addition of growth medium equivalent to 10-percent of the stock medium concentration. The elutriate and elutriate to disposal site water mixtures were repeated for each sediment sampling site within a location.

Four replicates of each treatment level and each control were established. The flasks were randomly distributed in two psychrotherm incubators (New Brunswick Scientific Co., Inc.). The temperature was 18°C for marine algal assays and 24°C for freshwater assays (± 2°C). Cool-white fluorescent bulbs were used to obtain constant illumination of approximately 1100 to 1300 µw/cm^2. The shaking rate was 110 rpm

throughout the assays. Growth was followed for 14 days in the Ashtabula samples and for 8 days in the Houston Ship Channel samples.

The inoculum was prepared by centrifuging and washing stock culture cells with sterile water containing 15 mg $NaHCO_3$ per litre for the freshwater algae or with sterile artificial seawater without nutrients for the marine algae. The inoculum cell concentration was adjusted by dilution, then pipetted into the test water to give a starting concentration in the test waters of 10^3 cells per millilitre for S. capricornutum and 10^2 cells per millilitre for D. tertiolecta.

Growth of the test organisms, as measured by total cell numbers, was used to measure the response of the organisms. Growth in disposal site water was considered as baseline and growth in the various dilutions of elutriate was compared to the baseline. Maximum standing crop (maximum number of cells) under each test condition was the variable of interest. Cell numbers were determined using a Coulter Electronic Particle Counter Model TA II.

Statistical analyses of the data included Duncan's New Multiple Range tests, analyses of variance, and T-tests.

ASHTABULA HARBOR RESULTS

Physical Characteristics of the Samples and Elutriates

Predredging samples were collected from the harbor and disposal site on 31 July 1975, four days before the dredging operation began. Sediment and water samples for preparation of the elutriates were taken from two areas within the harbor, one site designated polluted (Site P) and the other site designated unpolluted (Site UP) by the EPA. Composite water column samples were taken from a central location within the disposal area by collecting water samples 1 m from the bottom, at mid-depth in water column, and 1 m below the water surface. Dredge site water was collected 1 m above the sediment surface.

Samples were collected during dredging from the dredge hopper at each of the two dredging sites and during disposal from each plume as

disposal was occurring. There were two dump sites within the disposal area, one for the polluted dredged material and one for the nonpolluted dredged material. Table 1 lists the results of field measurements taken during the sampling period. The only significant difference is the lower dissolved oxygen concentration at Site P.

Table 2 lists the pH of the samples used for algal studies before and after growth. All samples had a higher pH after algal growth. The starting pH of Site P elutriate was 7.0 while that of Site UP was 8.1. The pH difference could have contributed to the variation in growth between elutriates.

Chemical Analysis Before Algal Growth

Table 3 lists the chemical analyses of the Ashtabula and Lake Erie samples before they were used for the algal growth studies. Only the values for ammonia plus ammonium ($NH_3+NH_4^+$), manganese, and iron are shown. These were the only constituents that had a significant change in concentration between disposal site water and elutriates. Sediments from both sites released $NH_3+NH_4^+$ into the elutriate with Site P releasing about twice the amount released from Site UP. Both hopper slurries also contained $NH_3+NH_4^+$ in high concentrations, but these constituents were not detectable in either disposal plume. Manganese was released from both sediment sites and was found in the hopper slurry from both sites, but was not detectable in either disposal plume. Iron was removed from the dredge site water during the preparation of both elutriates. High concentrations of iron were found in the hopper slurries and disposal plumes from both sediment sites.

Algal Growth

Table 4 lists the maximum cell yield for Selenastrum capricornutum under various conditions of growth. Maximum cell yield in 100-percent-disposal-site water was approximately 9.20×10^3 cells per ml. When nutrients were added to the disposal site water, the growth yield increased to 3.34×10^5 cells per ml. Elutriate from the site designated polluted produced an inhibitory effect with a maximum yield of 700×10^3. Nutrient addition did not significantly increase the yield. Increased growth did not occur when nutrients were added to the

combination of 50 percent elutriate to 50 percent disposal site water. The fact that nutrient additions did not stimulate growth in Site P elutriate would indicate the presence of a toxic substance.

The site designated as unpolluted also had an inhibitory effect. Growth yield in 100-percent elutriate from Site UP was 7.10×10^3 cells per millilitre. When nutrients were added, the yield increased to 1.91×10^4 per millilitres. Cell yield in 50-percent disposal site water to 50-percent elutriate from Site UP was 5.60×10^3; adding nutrients increased the yield to 2.08×10^4 cells per millilitre. Adding nutrients to 100-percent disposal site water resulted in a large increase in the cell yield. This was not the case for the elutriate preparation. Therefore, both sediments used in this study released toxic substances, but Site P apparently released more than Site UP.

Table 4 also lists the results of algal growth in the hopper slurries and disposal plumes from each site. Growth in Site P hopper slurry (4,600 cells per millilitre) was much less than in the elutriate (7,000 cells per millilitre) while growth in the disposal plume (8,500 cells per millilitre) was greater than growth in the elutriate. Nutrient additions increased growth in the slurry and plume to approximately 400,000 cells per millilitre.

Maximum cell yield in the hopper slurry and disposal plume for Site UP was 6,600 and 8,800 cells per millilitre, respectively. Nutrient additions increased the cell yield to 260,000 per millilitre in the hopper slurry and 560,000 in the disposal plume samples.

To summarize the algal growth data, there was less growth in the elutriates than in the disposal site water. Less growth occurred in the hopper slurry than in the elutriate from Site P; while growth in the plume sample was greater than elutriate growth from this site. The elutriate and hopper sample had approximately the same yield for Site UP, but growth was higher in the plume sample than in the elutriate.

Nutrient spikes increased the growth yield in all cases reported. The magnitude of the increase was much greater in the case of the hopper slurry and plume samples than in the case of elutriate from either site. This may indicate that the elutriate is a "worst case" measurement of

toxicants released from the sediment.

Chemical Analysis After Algal Growth

Table 5 lists the concentration of NH_3+NH_4^+, manganese, and iron remaining after the growth experiments. Ammonia plus ammonium was reduced to below detectable limits in both elutriates, while it was decreased only 12.5 percent in the hopper slurry of Site P and 64 percent in the hopper slurry from Site UP. Manganese decreased 47 percent in the elutriate and was almost gone from the hopper slurry of Site P. It decreased 36 percent in the elutriate and 60 percent in the hopper slurry of Site UP. Essentially all of the iron was gone from all of the samples.

HOUSTON SHIP CHANNEL RESULTS

Physical Characteristics of the Samples and Elutriates

Samples were collected 23 September 1975. Disposal site water was collected at mile 0 (Morgan's Point) of the Houston Ship Channel. Mile 0 was also used as sediment collection Site 1. Mile 16 was used as sediment collection Site 2. Strong currents prevented collection of sediments at Site 2 from the center of the channel, so samples were collected on the side of the channel in 1.5 m of water. Table 6 lists field measurements taken during sampling. Site 2 had a much lower dissolved oxygen concentration and salinity than Site 1.

Table 7 lists the pH and salinity of the samples used for algal growth studies. The pH rose slightly in both elutriates, but decreased slightly in the disposal site water. Since dredge Site 2 had a lower salinity than Site 1, disposal site water was used to prepare the elutriate for Site 2. The salinity for disposal site water and both elutriates did not change as a result of algal growth.

Chemical Analysis Before Algal Growth

Table 8 lists the chemical analysis of Houston Ship Channel water samples and elutriates before algal bioassays. Ammonia plus ammonium-nitrogen (NH_3+NH_4^+) were released from the sediments. Disposal and dredge site water had high levels of orthophosphate, a high percentage

of which was adsorbed by the sediments during elutriate preparation. Significant quantities of total organic and inorganic carbon were released during preparation of the elutriate. Of the heavy metals analyzed, only manganese was released from the sediments. The concentration of iron was high in all samples analyzed.

Algal growth

Table 9 lists the results of growth experiments using Dunaliella tertiolecta in disposal site water and elutriates prepared from sediment Sites 1 and 2 of the Houston Ship Channel. Growth was better in all concentrations of elutriate and disposal site water than it was in 100-percent disposal site water. As the elutriate concentration was increased, the maximum cell yield decreased. In 100-percent disposal site water, the maximum cell yield was 0.25×10^6 cells per millilitre. In 25-percent elutriate to 75-percent disposal site water from Site 1, the yield increased to 1.57×10^6 cells per millilitre. The cell yield decreased as the elutriate concentration was increased, resulting in a cell yield of 0.95×10^6 for 100-percent elutriate of Site 1.

The same trend occurred for sediment Site 2 although the maximum cell yield was different for each mixture when compared to Site 1. Maximum cell yield in 25-percent elutriate to 75-percent disposal site water was 1.99×10^6 cells per millilitre and decreased as the elutriate was added; the cell yield in 100-percent elutriate was only 0.41×10^6 cells per millilitre.

Nutrient additions increased the cell yield in all cases, but the magnitude of the increase was greater in 100-percent disposal site water than in 100-percent elutriate from either site. Toxic substances were apparently released from the sediments with the result that as the elutriate concentration was increased, the toxic substances inhibited algal growth.

Chemical Analysis After Algal Growth

Table 10 lists the concentrations of nutrients and heavy metals remaining after algal growth in 100-percent disposal site water and 100-percent elutriate from Sites 1 and 2. The NH_3+NH_4^+ concentration decreased from 10.0 to 0.4 ppm in Site 1 elutriate, while it only

decreased from 17.0 to 13.5 ppm in Site 2 elutriate. Orthophosphate decreased by approximately 50-percent in the disposal site water but was essentially the same in both elutriates.

Manganese decreased in both elutriates by approximately the same relative amount even though Site 1 elutriate had a much higher concentration than Site 2. Iron decreased by approximately the same amount in the disposal site water and elutriates. The slight increase in some of the heavy metal concentrations can be attributed to nutrient carryover since the algal medium contains zinc and copper.

CONCLUSIONS

Algal bioassays are useful in evaluating the potential biological effects of the chemical constituents released from the sediment on photoplankton at the disposal site. Stimulation, as well as toxicity, of algal growth has been observed in initial bioassays.

During a dredged material disposal operation, the contaminants released from the sediment are present in the water column for a short period of time and cannot be detected a few seconds after disposal operations cease. The algal population that is in the water column and exposed to the contaminants is reproducing at a rapid rate in comparison to many other water column organisms. Therefore, algae may remove these contaminants at a rapid rate relative to other organisms and serve as an important entrance of chemical compounds, including toxicants, into the aquatic food web.

Algae grow and reproduce at a rapid rate, are easily worked with, can be maintained in a small amount of space, and require no expensive or complicated equipment. Therefore, algal bioassays are a rapid, simple method for routine screening of potential toxicants released from the sediment.

There are two approaches that can be used in applying bioassay data to determine the acceptability of a particular dredged material for disposal. The first method would involve comparing growth in 100 percent elutriate and 100 percent disposal site water. The effect of diluting the elutriate with disposal site water would be considered.

The second approach would involve growth of the test organism in elutriates which have been characterized for major chemical constituents and would attempt to compare the biological response of the test organisms to the concentrations of various nutrients and heavy metals found in the elutriates. State and Federal water-quality standards, as well as published literature, could be used in evaluating the data. The method could help establish criteria or standards for disposal of dredged material. It suffers from the fact that a chemical constituent not included in the chemical analyses may have caused the effect. Also, there is a lack of knowledge as to the biological response caused by a mixture of chemicals (e.g., synergistic effects of heavy metals).

In relation to the first approach, algal bioassays of the elutriate would indicate the bioavailability of dissolved constituents released from dredged material and the possible effect on phytoplankton productivity at the disposal site. If observed growth in the elutriate were equivalent to observed growth in disposal site water, it would indicate that no adverse effect on the phytoplankton would be expected at the disposal site. If a stimulatory or inhibitory response were observed in the elutriate cultures, mixing and diffusion at the disposal site must be considered in evaluating the bioassay results. The procedure described used various ratios of elutriate and disposal site water in an attempt to simulate dilution. Duration of exposure to a particular elutriate concentration was not considered and each dilution was considered a "worst case" situation. The various dilutions used were considered arbitrary and more appropriate dilutions could be substituted as needed.

The EPA has published proposed criteria of freshwater quality for aquatic life (13). The "Maximum Acceptable" level for most constituents is based on bioassays using the most sensitive species in the locality as a test organism and the receiving water as the test matrix. For example, the concentration of un-ionized ammonia should not exceed 0.05 times the concentration which is lethal to 50 percent of the test organisms in 96 hr [0.05 (96 hr LC_{50})]. They also suggest an "Unacceptable" concentration for various constituents and for un-ionized

ammonia; it is suggested that the concentration be less than 0.02 mg per litre.

In relation to the Ashtabula chemical analyses (Table 3), the concentration of un-ionized ammonia at pH 7.0 is about 1.4 percent (16) of the reported value for Site P elutriate or 0.14 ppm. At pH 8.0 the concentration is approximately 3.5 percent of the reported values for Site UP which was equal to 0.21 ppm. The hopper slurries contained 0.28 ppm and 0.38 ppm for Sites P and UP, respectively. Each of these concentrations exceeds the "Unacceptable" level for ammonia. The proposed criteria do not list the concentrations of iron and manganese that are unacceptable for freshwater aquatic life.

Rachlin and Farran (14) found that the growth of the green algal, Chlorella vulgaris, was reduced approximately 50 percent in the presence of 2.0 ppm zinc. Payne (12) reported that in waters not containing chelating agents, the toxic level of zinc was 45 ppb for Selenastrum capricornutum. The highest concentration of zinc found in the Ashtabula elutriates was 20 ppb. It is interesting to note that the Algal Assay Procedure growth medium contained 15-ppb zinc.

The EPA also has published proposed criteria for marine water quality for aquatic life. The "Maximum Acceptable" concentrations are based on bioassays as previously discussed. "Unacceptable" levels are also listed. For marine water, it is unacceptable for the concentration of ammonia to exceed 0.4 ppm. Using the concentrations of $NH_3+NH_4^+$ listed in Table 8 to calculate the ammonia values for the Houston samples, the dredge site waters contained 0.01 and 0.03 ppm for Sites 1 and 2, respectively. The elutriate from Site 1 contained 0.35 ppm while Site 2 elutriate exceeded the unacceptable level since it had a concentration of 0.59 ppm.

Of the heavy metals listed in Table 8, iron and manganese exceeded the suggested unacceptable levels. Iron exceeded the proposed level of 300 ppb in all of the Houston samples. Manganese exceeded the suggested level of 100 ppb in both elutriates. The heavy metal concentrations found in the elutriates indicated a potential water quality problem. The bioassay data shown in Table 9 also indicated a potential problem in

relation to phytoplankton productivity caused by contaminants released from the sediment.

Erickson, et al. (5) have shown that a concentration of 450-ppb copper inhibited the growth of D. tertiolecta by 50 percent of that observed in the controls. Overnell (11) inhibited the photosynthetic oxygen evolution of D. tertiolecta by 50 percent in the presence of 640-ppb copper. The toxic level reported by Kemp et al. (8) for eight species of green algae was 2.0-ppm copper. The maximum concentration of copper found in the elutriates was 27 ppb, far less than any of the reported values that caused toxic effects.

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TABLE 1. FIELD MEASUREMENTS TAKEN DURING SAMPLE COLLECTION AT THE LAKE ERIE DISPOSAL SITE AND IN ASHTABULA HARBOR

Sample	Depth in Metres	Dissolved Oxygen in Parts per Million	Temperature in Degrees Centigrade
Disposal-site water	0	9.6	17.0
	3	8.7	18.5
	6	8.7	19.0
	9	8.0	19.0
	12	8.0	19.0
	15	8.0	19.0
Site P water	0	8.6	28.0
	3	5.5	25.0
	6	3.1	24.0
Site UP water	0	9.9	27.0
	3	9.5	25.0
	6	6.8	24.5
	8	6.7	24.0

TABLE 2. LABORATORY MEASUREMENTS OF pH ON ASHTABULA SAMPLES USED FOR GROWTH STUDIES OF *Selenastrum capricornutum*

Sample	pH	
	Before Algal Growth	After Algal Growth
Disposal site water	7.2	8.2
Dredge site P	7.6	-
Dredge site UP	7.9	-
Elutriate site P	7.0	8.5
Elutriate site UP	8.1	8.3
Hopper slurry site P	7.9	8.0
Hopper slurry site UP	8.0	8.0
Disposal plume site P	8.2	8.3
Disposal plume site UP	8.3	8.4

TABLE 3. CHEMICAL ANALYSIS OF LAKE DISPOSAL SITE WATER, ASHTABULA HARBOR DREDGE SITE WATERS, ELUTRIATES, HOPPER SLURRIES, AND DISPOSAL PLUMES BEFORE ALGAL BIOASSAYS

Sample	Constituent		
	Ammonia Plus Ammonium in Parts Per Million	Manganese in Parts Per Billion	Iron in Parts Per Billion
Disposal-site water	<0.1	5	20
Site P			
Dredge-site water	0.2	98	520
Elutriate	11.0	750	50
Hopper slurry	8.0	614	93,500
Disposal plume	<0.1	<1	4,000
Site UP			
Dredge-site water	<0.1	29	230
Elutriate	6.0	700	14
Hopper slurry	11.0	650	25,000
Disposal plume	<0.1	<1	25,000

TABLE 4. MAXIMUM GROWTH OF *Selenastrum capricornutum* IN LAKE ERIE DISPOSAL SITE WATER AND ASHTABULA HARBOR ELUTRIATES, HOPPER SLURRIES, AND DISPOSAL PLUMES

Growth Condition	Average Maximum Standing Crop in Cells Per Militre	
	Without Spike	With Spike
100% Disposal site water	9,200 ± 970	334,000 ±38,000
Site P		
100% Elutriate	7,000 ± 1,120	8,700 ± 1,510
50% Elutriate:50% Disposal site water	8,000 ± 1,410	8,200 ± 1,260
Hopper slurry	4,600 ± 930	400,000 ±31,000
Disposal plume	8,500 ± 570	404,000 ±40,000
Site UP		
100% Elutriate	7,000 ± 1,920	19,100 ± 3,060
50% Elutriate:50% Disposal site water	5,600 ± 1,790	20,800 ± 3,130
Hopper slurry	6,600 ± 300	260,000 ±14,000
Disposal plume	8,800 ± 700	560,000 ±61,000
100% Growth medium	4,000,000	- -
10% Growth medium	598,000 ± 59,000	- -

TABLE 5. CHEMICAL ANALYSIS OF LAKE ERIE DISPOSAL SITE WATER AND ASHTABULA HARBOR ELUTRIATES, HOPPER SLURRIES, AND DISPOSAL PLUMES AFTER ALGAL BIOASSAYS

Sample	Constituent		
	Ammonia Plus Ammonium in Parts Per Million	Manganese in Parts Per Billion	Iron in Parts Per Billion
Disposal site water	<0.05	<1	5
Site P			
Elutriate	<0.05	400	3
Hopper slurry	7.0	6	3
Disposal plume	0.2	<1	4
Site UP			
Elutriate	<0.05	450	25
Hopper slurry	4.0	280	2
Disposal plume	0.2	<1	9

TABLE 6. PHYSICAL MEASUREMENTS TAKEN DURING SAMPLE COLLECTION IN THE HOUSTON SHIP CHANNEL

Sample	Depth in Metres	Dissolved Oxygen in Parts per Million	Temperature in Degrees Centigrade	Salinity in Parts per Thousand
Disposal site water	3	8.0	24.0	14.0
Site 1 water	0	8.3	24.0	14.0
	3	8.0	24.0	14.0
	6	-	24.0	13.5
	9	-	24.0	14.0
	12	7.3	24.0	14.0
Site 2 water	0	1.4	26.5	6.0
	3	1.2	27.0	6.5
	6	0.9	27.0	6.5
	9	1.2	27.0	7.0
	12	1.8	27.0	7.5

TABLE 7. LABORATORY MEASUREMENTS OF pH AND SALINITY ON HOUSTON SHIP CHANNEL SAMPLES USED FOR GROWTH STUDIES OF *Dunaliella tertiolecta*

Sample	Before Algal Growth		After Algal Growth	
	pH	Salinity in Parts per Thousand	pH	Salinity in Parts per Thousand
Disposal site water	8.2	15	8.0	15
Dredge site 1 water	8.2	15	8.3	-
Dredge site 2 water	8.2	8	8.3	-
Elutriate site 1	8.4	16	8.3	16
Elutriate site 2	8.4	15	8.3	15

TABLE 8. CHEMICAL ANALYSIS OF HOUSTON SHIP CHANNEL SAMPLES AND ELUTRIATES BEFORE ALGAL BIOASSAYS

Constituent	Disposal Site Water	Dredge Site Water		Elutriate	
		Site 1	Site 2	Site 1	Site 2
Nutrients (ppm):					
NO_3	0.2	0.2	0.8	0.4	0.4
NO_2	0.1	0.1	0.8	0.02	0.3
NH_3+NH_4	0.2	0.3	1.0	10.0	17.0
TKN	<1.0	<1.0	1.0	17.0	17.0
OPO_4	1.3	1.2	2.1	0.5	0.3
$AHPO_4$	1.3	1.2	2.1	0.5	0.3
TIC	11.0	16.0	22.0	34.0	39.0
TOC	19.0	16.0	16.0	29.0	26.0
Heavy metals (ppb):					
Cd	2	1	2	2	1
Ni	5	5	7	5	2
Zn	10	<10	<10	10	<10
Mn	45	40	40	8000	1000
Cu	9	6	5	4	4
Fe	4000	4000	4500	4100	4600
As	4	4	6	3	2

TABLE 9. MAXIMUM GROWTH OF *Dunaliella tertiolecta* IN HOUSTON SHIP CHANNEL SAMPLES

Growth Conditions	Average Maximum Standing Crop	
	Without Spike ($\times 10^6$ Cells Per Millilitre)	With Spike ($\times 10^6$ Cells Per Millilitre)
100% Disposal site water	0.25 ± 0.017	0.56 ± 0.021
Site 1		
100% Elutriate	0.95 ± 0.075	1.08 ± 0.035
75% Elutriate:25% Disposal site water	1.29 ± 0.016	- -
50% Elutriate:50% Disposal site water	1.59 ± 0.062	1.65 ± 0.014
25% Elutriate:75% Disposal site water	1.57 ± 0.005	- -
Site 2		
100% Elutriate	0.41 ± 0.042	0.54 ± 0.008
75% Elutriate:25% Disposal site water	0.84 ± 0.035	- -
50% Elutriate:50% Disposal site water	1.35 ± 0.030	1.51 ± 0.086
25% Elutriate:75% Disposal site water	1.99 ± 0.063	- -
100% Growth medium	3.60	-
10% Growth medium	0.20 ± 0.014	-

TABLE 10. CHEMICAL ANALYSIS OF HOUSTON SHIP CHANNEL SAMPLES AND ELUTRIATES AFTER ALGAL BIOASSAYS

Constituent	Disposal Site Water	Elutriate Site 1	Elutriate Site 2
Nutrients (ppm):			
NO_3	0.005	0.1	0.4
NO_2	<0.005	0.005	0.01
NH_4-NH_3	0.2	0.4	13.5
TKN	<1.0	2.0	22.7
OPO_4	0.7	0.5	0.3
$AHPO_4$	0.7	0.5	0.3
TIC	20.0	33.0	30.0
TOC	14.0	38.0	33.0
Heavy metals (ppb):			
Cd	<2	3	2
Ni	9	9	38
Zn	30	37	39
Mn	3900	7300	540
Cu	7	27	20
Fe	2850	2640	2750
As	2	<2	<2

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Shuba, Peter J

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